genome are targeted and incorporated into progeny virus particles. The viral genome consists of eight negative strand RNA segments which are tightly packed by the nucleoprotein (NP) forming ribonucleoprotein complexes (RNPs). They are enclosed by a layer of matrix protein M1 and the viral membrane.
In vitro, we have studied the interaction of viral RNPs and the matrix protein M1 with large unilamellar vesicles of various lipid compositions by flotation assay and found that vRNPs alone are not able to associate with model lipid membranes. However, our findings suggest that M1 is able to mediate the binding of vRNPs to lipid bilayers.
In a new approach focusing on NP in a cellular context, we investigate the intrinsic properties of this protein essential for transport and targeting to the budding site. Fusion constructs of NP with fluorescent proteins are used to determine intracellular localization and the photoactivatable fluorescent protein Dendra2 allows us to investigate the dynamics of NP in different cellular compartments in living cells. Intracellular localization of tagged NP is very similar to that of wildtype NP. Hence, tracking of fluorescently tagged NP in virus infected cells is an interesting tool to study pathway and kinetics of intracellular transport of the viral RNP complexes during an infection cycle.

3408-Pos
Effects of Salts on Internal DNA Pressure and Mechanical Stability of Phages
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Our recent nanoindentation measurements on phage lambda, revealed an evolutionary optimization of DNA density in viral capsid. Based on these experimental data, we proposed that water hydrating DNA in the capsid, provides significant support against external capsid deformation at wild-type DNA packing density. Shorter DNA length mutants are on the other hand two times weaker just like empty capsids. In this work, we perform a stringent test of this assumption. DNA hydration force can be dramatically decreased by addition of multivalent ions (here Mg2+ and Sp4+). Indeed, AFM measurements demonstrate that spring constant for wt-DNA phage lambda decreases to a value of an empty capsid upon addition of multivalent salt compared to the ''zero-added-salt'' value obtained in the previous work. This data is systematically analyzed with DNA hydration model and further comparison is made with phage fi29.

3409-Pos
Role of the Electrostatic Interactions in the Genome Packaging and Ejection of DNA From Bacteriophages
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Electrostatic interactions play an important role in both packaging of DNA inside bacteriophages and its release into bacterial cells. While at the physiological conditions DNA strands repel each other, the presence of polyvalent cations such as spermine and spermidine in DNA solutions leads to the formation of DNA condensates. This phenomenon has been experimentally observed for DNA confined inside bacteriophages and upon its ejection into bacteria. In this presentation, we discuss packaging and release of DNA from bacteriophages under repulsive and attractive conditions using a coarse-grained model of DNA and capsids. The first group of simulations describes packaging of DNA inside bacteriophages Lambda. Packaging under repulsive conditions leads to the appearance of the folded toroidal conformations; DNA occupies all available space inside the capsid. Under the attractive potential both packed systems reveal toroidal conformations, leaving the central part of the capsids unoccupied by DNA. We also present a detailed thermodynamic analysis of packaging and show that the forces required to pack the genomes in the presence of polyamines are significantly lower than those observed under repulsive conditions (in the absence of polycations). Additionally we report the results of simulations of DNA condensation inside partially packed bacteriophage Lambda. In the second group of studies we simulated the ejection of DNA from bacteriophages. Simulations performed in the repulsive regime result in the formation of a random coil of fully ejected DNA, while the genome condenses into rod-like structures upon ejection, if the simulations were done with the attractive potential. In both cases we confirm the ''push-pull'' mechanism proposed to explain the ejection and estimate the pulling force that acts on the ejected portion of DNA.

3410-Pos
The Entropic Penalty of Confining a Chain Polymer into a Very Small Space
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The confinement of a flexible polymer is thermodynamically unfavorable, because of the reduction in the number of conformational states. The determination of the entropic penalty of confinement into a very small space is an important unsolved problem in polymer statistical mechanics. We present a method for calculating TΔS for the confinement of an elastic polymer of persistence length P when volume exclusion effects are ignored, considering three geometries: (1) parallel planes separated by a distance d; (2) a circular tube of diameter d; and (3) a sphere of diameter d. As d/P drops from 100 to 0.01, TΔS rises from about 0.01kT to about 30kT for both cases (1) and (2), with the cost in the latter case being consistently about twice that for confinement between parallel planes. The entropic penalty for confinement to a sphere is ~5kT per persistence length, when d = P, in the absence of excluded volume effects. TΔS can be determined fairly easily when chains of finite diameter are confined into thin tubes, or into spheres with diameters on the order of the persistence length. We also show how volume exclusion effects can be determined in other cases. Excluded volume effects can be very large, especially for confinement to spheres.

3411-Pos
Toward Understanding the Effect of Single Amino Acid Mutations on Viral Capsid Stability
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We are using the tools of computational biophysics to understand the mechanisms of adaptive protein evolution in viruses. Previous experimental studies have shown that single amino acid mutations in bacteriophage (virus that infect bacteria) ID11 result in large fitness (population doublings per hour) increases. These mutations occur near protein-protein interfaces motivating our hypothesis that these mutations increase the stability of the viral capsid. We used computer simulation to calculate the protein-protein binding affinity changes due to single amino acid mutations. We present these results that directly estimate the stability of the capsid. Due to the large size of the capsid, we explicity simulated atoms within a spherical region centered on the mutation with all other atoms held stationary. Our results show that the mutants have lower binding affinity than the ancestor, i.e., the mutant viral capsid is more stable. We also discuss capsid stability as a possible evolutionary mechanism.

3412-Pos
Respiratory Syncytial Virus Interactions with Nanoparticles Using Transmission Electron Microscopy
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Respiratory Syncytial Virus is the leading cause of lower respiratory tract infections in infants and children worldwide, with almost all children becoming infected by the age of 2 years. It is leading cause of bronchiolitis, pneumonia, mechanical ventilation, and respiratory failure in infants in the US. Nanoparticles have been gaining usage in medicine and biological application due to their size and other properties. Few studies have been done in their use as therapy. Silver nanoparticles have been shown to interact with surface protein of HIV virus. In the present study, we studied the interaction of nanoparticles with RSV using Transmission electron microscopy (TEM). RSV has surface proteins F and G which are essential for RSV infection to host cells. Interaction or attachment of nanoparticles to the surface proteins of RSV opens up the possibility of preventing RSV infection to host cells. Human cell lines were infected with RSV and RSV incubated with nanoparticles for different time intervals. Samples were negatively stained and analysed using TEM. TEM studies showed RSV to be polymorphic with size ranging from 80-150 nm. Our initial results also indicate binding of nanoparticles (silver and gold) to RSV surface mainly the proteins present on RSV. Cells incubated with nanoparticles were also analyzed to determine endocytosis pathway. Ultrathin sections (5 nm) of the cells incubated with nanoparticles were cut and examined using TEM. Initial studies indicate presence of nanoparticles mainly in the vesicles of the cells. Work is currently on the way to determine the pathways of nanoparticle endocytosis by cell lines.

3413-Pos
Characterization of Retroviral Gag Behavior in the Cytoplasm of Living Cells Using Fluorescence Fluctuation Spectroscopy
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Retroviruses such as human immunodeficiency virus (HIV) and human T-cell leukemia virus (HTLV) have a huge impact on human health worldwide.